

Week 11 – Mutations and Disease Exercises

Question 0

Once more, for my teaching project, I will need you to fill out an online questionnaire. This is compulsory!

Go to the course (27008) pages on [campusnet](#) and click on the “Evaluation” menu item. There’s a questionnaire called “Mid-term evaluation” to be filled up.

Thank you ☺

Question 1

Describe the differences between “somatic” and “germ line” mutations

Somatic: from “adult” cells; passed on by mitosis. Not in reproduction cells; not passed to offspring

Germ Line: in reproductive cells; passed on to the offspring; eg: genetic diseases such as sickle cell anaemia, Huntington’s, etc

Question 2

Describe the difference between a “frame-shift” and a “non sense” mutation

A frame shift results in a different amino acid sequence as a base is added/removed thus changing the codon order

A nonsense mutation is caused by the appearance of a STOP codon in the sequence, resulting in the truncation of the gene product.

Question 3*

Which of the following is a

- A – Silent mutation **Mutation 3**
- B – Nonsense mutation **Mutation 1**
- C – Missense mutation **Mutation 2**

WILD TYPE: ATG GAG GGG TCA CAC GGC AAT AAC
MUTANT 1: ATG GAG GGG TAA CAC GGC AAT AAC
MUTANT 2: ATG GAC GGG TCA CAC GGC AAT AAC
MUTANT 3: ATG GAC GGT TCA CAC GGC AAT AAC

Question 4

Write the protein sequences resulting from the previous question

- 1- **Met Glu Gly Ser Gln His Gly Asn Asn**
- 2- **Met Glu Gly STOP**
- 3- **Met Asp Gly Ser Gln His Gly Asn Asn**
- 4- **Met Glu Gly Ser Gln His Gly Asn Asn**

Question 5

List and explain the main types of chromosomal mutations

Translocations - Translocations are the transfer of a piece of one chromosome to a non-homologous chromosome. They are often reciprocal, with the two chromosomes swapping segments with each other. An abnormal hybrid gene is created leading to the production of a novel protein that is not normally found in the cell.

Inversion - A region of DNA on the chromosome can flip its orientation with respect to the rest of the chromosome.

Deletions - A large section of a chromosome can be deleted resulting in the loss of a number of genes.

Duplications - In this mutation, some genes are duplicated and displayed twice on the same chromosome.

Chromosome non-disjunction - During cell division, the chromosomes fail to successfully separate to opposite poles, resulting in one of the daughter cells having an extra chromosome and the other daughter cell lacking one.

Question 6*

We have seen that 5-methylcytosine is a mutation hotspot. Explain why and how is the cell dealing with it.

Demethylation of 5-methylcytosine results in a C to T substitution, leading to a C->G to T->A mutation in the DNA sequence. Contrary to the case of old C deamination (C->U), this kind of mutation remains embedded in the genome and will propagate to the next cellular generations.

Question 7

What are restriction enzymes?

Restriction enzymes are a special class of Enzymes from bacteria that cut DNA at specific sequences.

Question 8

Why do bacteria use these enzymes?

Bacteria use these enzymes as a defence mechanisms against viral infections, as they can destroy a foreign DNA by chopping it up to pieces.

Question 9

Why are these enzymes so important in biology?

Restriction enzymes have found a vast number of applications in research and in forensic medicine and diagnostics. They allow for DNA manipulation in the lab for the purpose of protein cloning, DNA identification, paternity tests, genetic screening and many more.

Question 10

How do bacteria avoid cutting their own DNA?

Bacterial DNA is heavily methylated at their C-bases. These methyl groups are able to block the action of restriction enzymes.

Question 11*

Describe DNA gel electrophoresis. What kind of information do we get from DNA gel electrophoresis?

- A technique for separating molecules (such as DNA fragments) from one another on the basis of their electric charges and molecular weights by applying an electric field to a gel.

- Number of fragments; size of fragments; relative amounts of fragments;

Question 12*

What is the particularity of TSE (aka Mad Cow Disease)? How is the disease transmitted and how does it proliferate in an organism?

It is a still unclear protein mutation not involving amino acid substitutions. The main difference between the healthy and the pathogenic proteins is structural (see fig 15.14 in book).

The disease is transmitted by incorporation of a misfolded PrP into the organism (mainly by ingestion of contaminated food). This misfolded protein will somehow induce properly folded proteins to misfold, in a chain-reaction manner.

Question 13*

One of the markers for sickle cell anaemia is a GAG->GTG mutation at codon 6 of the β -globin gene.

1) What type of mutation is happening here?

Missense mutation

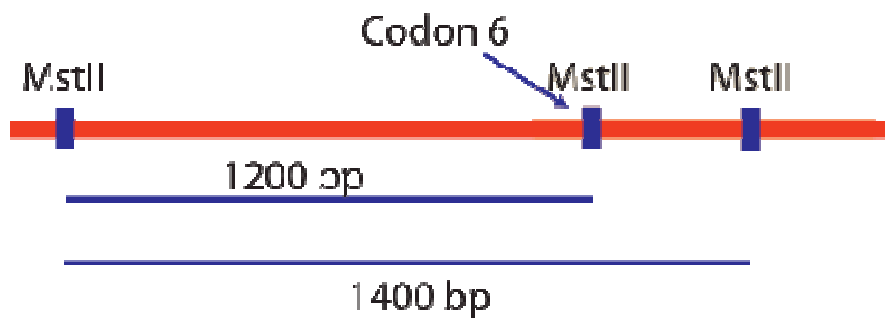
2) What amino acids are involved in the mutation (WT vs Mut)?

Glu -> Val

Question 14*

Both members of a newly wed couple are carriers for the sickle cell anaemia mutation. It is known that the mutation at codon 6 also results in the disruption of a MstII restriction site, as shown in the figure.

In order to determine whether the baby they just got is affected by the disease, a genetic screen by Restriction Fragment Length Polymorphism is performed on the parents and the newborn baby.



- 1) What is the background of this test?
- 2) How is it performed?
- 3) What results would you expect from the experiment? And why? (hint, remember Chapter 12)
- 4) What other method could you use to run the same test?

1) The background of the test is to use this difference in restriction sites to check whether a person's gene is WT or Mutant by testing the efficiency of the specific restriction enzyme on that DNA sequence.

2) The specific DNA sequence is isolated and incubated with the MstII enzyme and the correct buffers. After a certain time, the DNA containing the correct site will be cut into various fragments while the mutant gene will remain intact. Separation of the fragments by DNA agarose gel electrophoresis will allow the scientists to detect which gene version each family member will have.

3) Both parents will have 2 copies of the gene and are carriers. Therefore they will both have a copy of each allele (WT and Mut). Loss of the restriction site in one of the alleles will result in half of the genetic material being "resistant" to the MstII enzyme. Therefore we would expect that the analysis of both parent's DNA will give 1400, 1200 and 200 bp fragments. If the child inherits both copies of the mutant, he will only show the 1400 bp band. If he inherits one allele each, he will be also a carrier (1400, 1200, 200bp). If he only inherits the WT allele, he will only show a 1200 and a 200 bp fragment.

4) The same test can be run by DNA Gel blotting, even though it would be more inconvenient.

Question 15 (optional and for fun)

Go to this [site](#) and complete the exercise....

What sizes are the bands you obtain?

6000, 3500, 1500